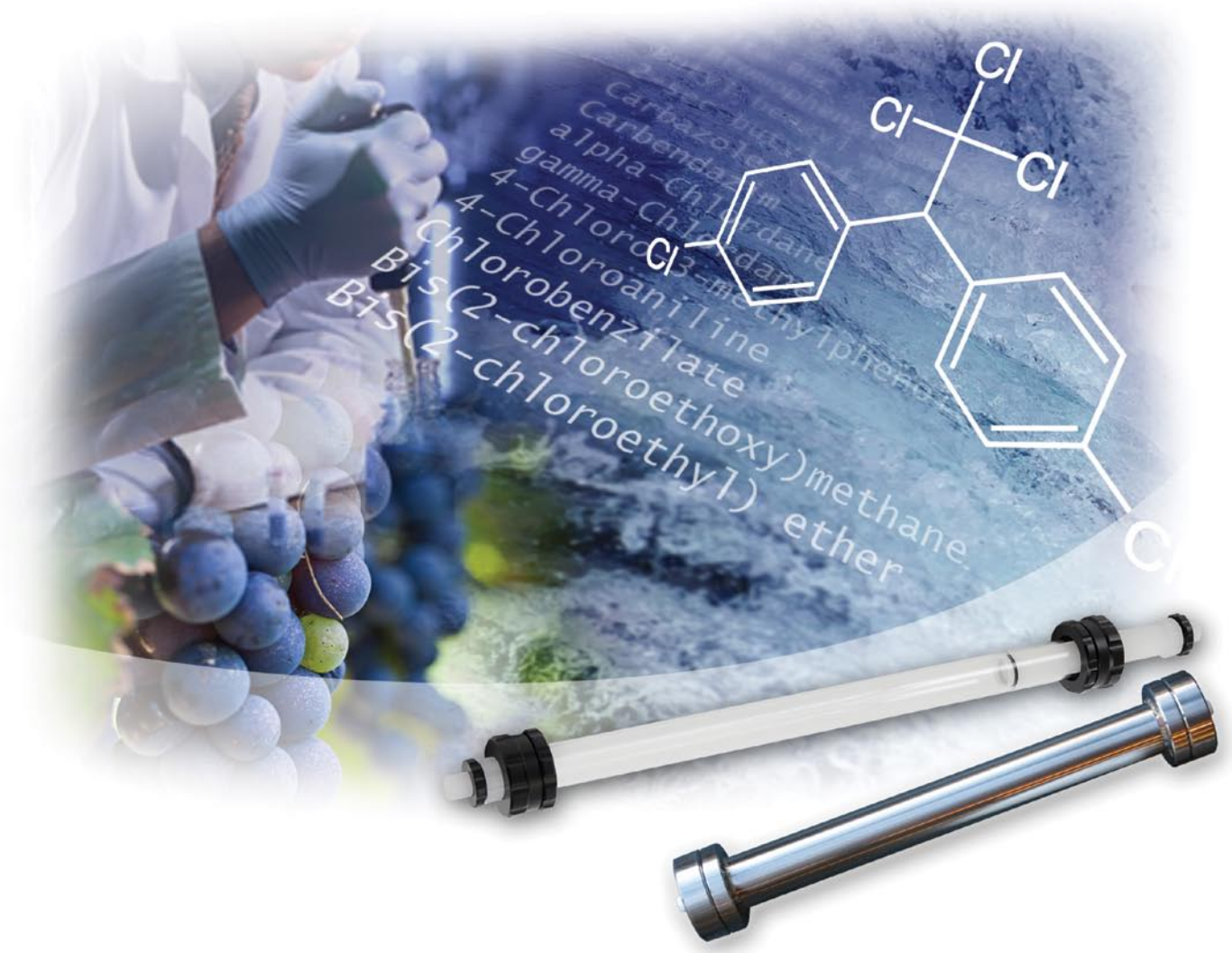
ENVIRONMENTAL
FOOD SAFETY

VARIAN, INC.

EcoSpheres™ and EnviroPrep™

SOLUTIONS FOR THE CLEAN-UP OF ENVIRONMENTAL & FOOD SAMPLES



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Agilent Technologies

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Sample Clean-up Solutions

Environmental & Food Safety Monitoring

Globally, government bodies require environmental and food samples to be analyzed for a wide range of contaminants. Many public and private bodies are charged with determining trace levels of pollutants from a wide range of biological sources. Common contaminants that are monitored in the environment and food include the many pesticides, herbicides and fungicides used in commercial farming.

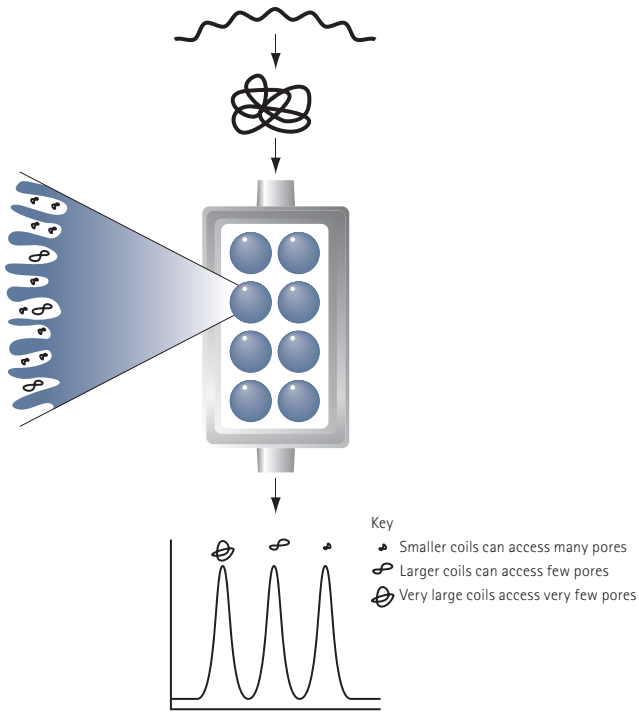
The analysis of pesticides and priority pollutants from complex environmental samples by GC, GC/MS, HPLC or LC/MS is more successful if low volatility, high molecular weight interferences, such as lipids, pigments, proteins, humic acids and plasticizers, are removed first. Failure to remove these interfering compounds can shorten column lifetime and increase analytical downtime.

Varian's Solution for Sample Clean-up by GPC

Using GPC, it is easy to isolate pesticides and priority pollutants from high molecular weight interferences, so that they can be collected for further analysis. The highly reproducible EcoSpheres™ and high resolution EnviroPrep™ products from Varian, Inc. allow samples from complex biological matrices, such as plant and animal tissue, to be cleaned up by GPC ready for further analysis of the contaminant.

The Benefits of Cleaning up Environmental Samples using EcoSpheres or EnviroPrep include:

- Removal of interferences which cause poor analytical results ensures a simple interpretation of the final analysis.
- Improved accuracy and linearity of the final analysis by the removal of extraneous samples gives lower detection limits and greater confidence in the results.
- A clean sample reduces the risk of damage to analytical equipment in the final analysis and can therefore reduce maintenance costs and downtime.
- Analysis of a cleaner sample improves the efficiency of the final analysis, leading to improved analysis quality and reduced analysis cost.
- EcoSpheres and EnviroPrep materials comply with guidelines in EPA method 3640A, DFG 519, EN1528, EN12393 and other regulatory bodies involved with food and environmental control.



Mechanism of Gel Permeation Chromatography/Size Exclusion Chromatography (GPC/SEC)

Clean-up by GPC

Gel permeation chromatography (GPC) is an ideal method for the clean-up of samples from environmental sources. A size-based separation is employed to separate the species of interest from higher molecular weight matrices. The size exclusion separation is based on partitioning of the components of the sample between a flowing liquid phase and stationary liquid trapped in the pores of a suitable material, typically porous crosslinked polymer beads. The pore size in the beads is carefully controlled such that smaller molecules are able to enter the pores, whereas large molecules have restricted access. Larger molecules therefore elute from the column very quickly whereas smaller molecules are retained and elute later.

Clean-up by GPC

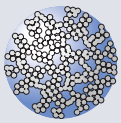
Microporous and Macroporous Materials for Clean-up

It is possible to use both microporous and macroporous materials for environmental and food clean-up by GPC. Varian, Inc. has introduced two materials for the clean-up of environmental samples.

The first is EcoSpheres, an economic, loose microporous material available in a dry powder form ready for swelling and gravity packing into glass columns. Microporous materials have low levels of crosslinking and no permanent pore structure; instead, pores are generated when these materials swell in solvents. Microporous packings allow higher loadings, where high resolution is not required.

The second is EnviroPrep, which are stainless steel columns packed with high resolution macroporous material ready for use on a preparative high pressure liquid chromatography system. Macroporous materials are highly crosslinked with a rigid and permanent pore structure which ensures high resolution separations, particularly where high sample loading is not required.

The GPC size exclusion separation and the properties of microporous and macroporous materials are shown below.

Product	Material		Performance	Swell	Mechanical Strength	Operating Conditions
EcoSpheres	Microporous		High loading, economic clean-up	High	Low	Low pressure
EnviroPrep	Macroporous		High resolution, high performance clean-up	Low	High	High pressure

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EcoSpheres

EcoSpheres

EcoSpheres beads are designed for the economic, low pressure separations of pesticides, chlorinated hydrocarbons and other small molecules from high molecular weight organic matrices.

EcoSpheres beads are supplied as loose media for packing into glass columns. They are microporous, nominally 45 µm, beads of styrene lightly crosslinked with divinyl benzene. The result is a product which is virtually free from fines and therefore highly reproducible, column after column, batch after batch.

Swelling in common organic solvents generates the pore structure of the EcoSpheres. When swollen, the material can be packed by gravity into glass columns.

Varian is also launching the EcoSpheres glass column hardware specifically designed to be leak-free and to avoid the issues of voiding and channeling, which can seriously affect the results obtained.

The new EcoSpheres glass columns are easy to pack, and once the column is packed and equilibrated, are very stable, leading to reliable data throughout the column lifetime.

Features and Benefits

- For faster analysis times, the large particle size of EcoSpheres beads allows a 25 mm ID glass column to be used at a flow rate of up to 10 mL/min.*
- For maximum flexibility, the EcoSpheres product can be used with any liquid chromatography system capable of isocratic flow at the required flow rates.
- The microporous EcoSpheres material maximizes loadings, is easy to use and delivers good quality resolution.
- Excellent sample peak shapes are obtained: the narrow particle size distribution of the EcoSpheres material ensures that the beads are easy to pack into columns.
- The EcoSpheres pore size has been selected to maximize the separation of low molecular weight species of interest from higher molecular weight interferences, which are excluded from the pores of the material.
- For flexibility, EcoSpheres beads can be packed into a variety of glass columns. Typical glass column dimensions are 300-450 x 25 mm.

* Maximum operational pressure 300 psi (21 bar)

Applications

The following applications highlight the performance of glass columns packed with the EcoSpheres material for sample clean-up applications.

EPA Test Separation

The EPA has defined a sample clean-up method for the removal of lipids and other high molecular weight interferences from a sample containing pesticides or other species of interest. As part of that method, a separation of a test solution is suggested to determine the performance of the separation. The GPC calibration solution is prepared in dichloromethane containing the following analytes (in elution order):

EPA Test Separation

Compound/KEY	mg/L
1. Corn Oil	25,000
2. Bis(2-ethylhexyl) phthalate	1,000
3. Methoxychlor	200
4. Perylene	20
5. Sulfur	80

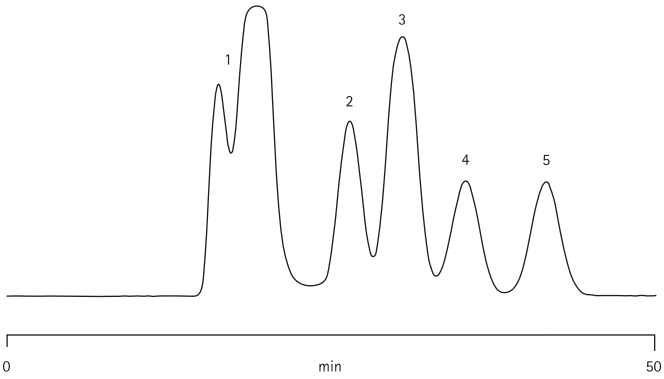

Figure 1. The criterion for the test separation is that each of the peaks can be resolved in the sample. Although the peaks are broad due to the use of a large particle size microporous packing material, EcoSpheres do resolve all of the components, illustrating their suitability for applications covered by the EPA method.

Column: EcoSpheres glass column, 450 x 25 mm
Eluent: Dichloromethane
Flow Rate: 5 mL/min
Detector: UV, 254 nm

Economical Sample Clean-up

Separation of Phthalate from Walnut Oil

Walnut oil, high quality oil known for its delicate, nutty flavor, is used in cooking. Walnut oil is an expensive commodity, produced mainly in France, that can sour if incorrectly stored. As part of a production process, walnut oil was to be packaged in plastic bottles manufactured from polyvinyl chloride (PVC). A stiff plastic, PVC is made flexible by the addition of plasticizers, typically phthalates, which help the polymer chains slide past each other. However, phthalates have been implicated in recent years as a potential health risk, raising concerns about phthalates leaching into foodstuffs.

This application shows the separation of a phthalate additive from a sample of walnut oil as part of a quality control procedure to isolate and measure the quantity and type of phthalate. To perform the separation, a 450 x 25 mm glass column was packed with EcoSpheres swollen in tetrahydrofuran.

Separation of Phthalate

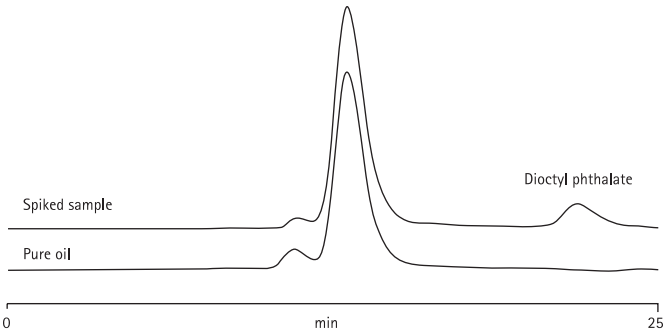

Figure 2. Clearly the phthalate is well resolved from the oil, allowing the small molecule to be isolated for further testing.

Column: EcoSpheres glass column, 450 x 25 mm
System: Modular system (dual piston pump with injection valve)
Inj Vol: 5 mL
Detector: UV at 254 nm
Solvent: Tetrahydrofuran
Flow Rate: 10 mL/min
Data collection/analysis: Cirrus GPC software and PL DataSteam data capture unit.

Accessories

Varian manufactures a wide range of accessories that may be used with the EcoSpheres products.

Ordering Information

Product	Part No.
EcoSpheres Loose Media, 100 g	PL1460-4M03
Complete Glass Column	PL1310-0054
Glass Column only (no end fittings)	PL1310-0055
Adjustable end piece	PL1310-0056
Fixed end piece	PL1310-0057
Seals (Kalrez)	PL1310-0058
Frits (glass, 10-13 µm)	PL1310-0059

VARIAN, INC.

EnviroPrep

EnviroPrep Packed Columns

- 10 µm particles for high resolution
- High pressure operation for flow rates of typically 5 mL/min
- Supplied in pre-packed stainless steel columns, ready for use
- 300 x 25 mm and 150 x 25 mm column formats - combine for maximum resolution
- 50 x 25 mm guard column - protect your EnviroPrep column against blockages

Features and Benefits

- For fast start up times and guaranteed performance, pre-packed EnviroPrep columns have been QC tested to ensure performance out of the box.
- For sample clean-up in minimal time with maximum throughput, highly mechanically stable EnviroPrep columns can be operated up to 2200 psi (150 bar) with eluent flows of up to 10 mL/min.
- For maximum flexibility, EnviroPrep columns can be used with any liquid chromatography system capable of isocratic flow at the required flow rates.
- To allow the separation of trace components at low concentrations, the rigid and well-defined pores of the macroporous packing material give unrivalled resolution and peak shape.
- The low pore size of EnviroPrep columns excludes high molecular material, maximizing separation of these interferences from the low molecular weight species of interest.

Applications

The following applications highlight the performance of the EnviroPrep columns in sample clean-up applications.

EPA Test Separation

The EPA has defined a sample clean-up method for the removal of lipids and other high molecular weight interferences from a sample containing pesticides or other species of interest. As part of that method, a separation of a test solution is suggested to determine the performance of the separation. The GPC calibration solution is prepared in dichloromethane containing the following analytes (in elution order):

EPA Test Separation

Compound/KEY	Approx. sample concentration / mg/mL
1. Corn Oil	25,000
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3. Methoxychlor	200
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5. Sulfur	80

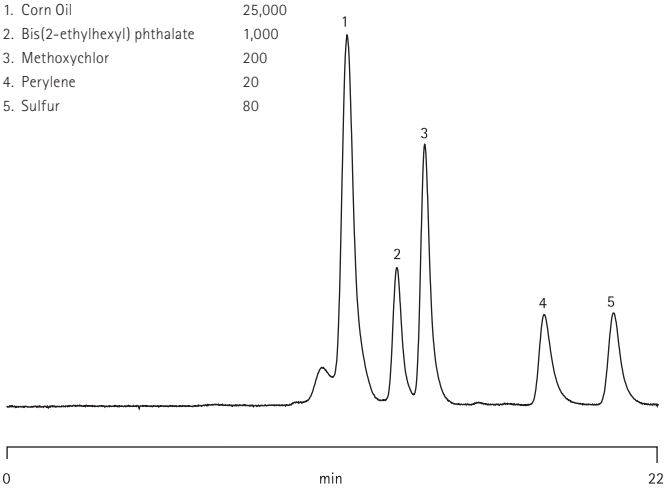

Figure 3. The criterion for the test separation is that each of the peaks can be resolved in the sample. With EnviroPrep, sharp peaks are obtained with clear separation, illustrating the use of the column for applications covered by the EPA method.

Column: 2xEnviroPrep, 300 x 25 mm (PL1210-6120EPA)
Eluent: Dichloromethane
Flow Rate: 10 mL/min
Detector: UV, 254 nm

High Resolution Sample Clean-up

Analysis of Pesticides in Mackerel

As part of a food safety program, the level of the pesticide 1,2,3,4,5,6-$C_6H_6Cl_6$ that was present in a sample of seafood was evaluated. A sample of mackerel was macerated and the tissue was extracted into chloroform. The resulting organic solution was then cleaned up by elution through two EnviroPrep 300 x 25 mm columns connected in series.

Pesticides in Mackerel

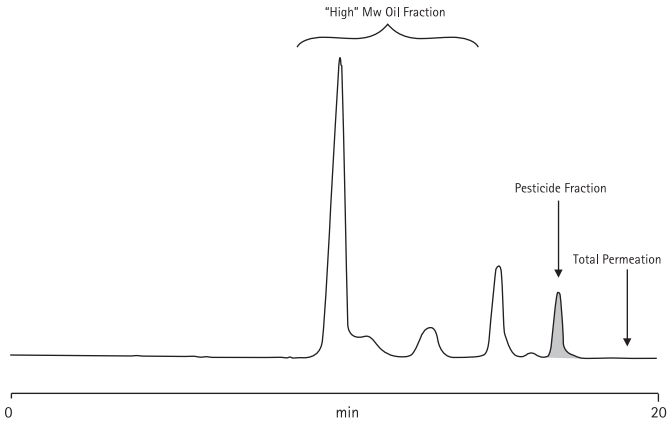

Figure 4. The chromatogram obtained clearly shows that the EnviroPrep columns can be used to separate high molecular weight lipids that are excluded on the EnviroPrep columns from a small molecule pesticide. This allowed collection of the pure pesticide in solvent for further analysis and demonstrated the use of EnviroPrep for cleaning up a complex sample.

Pesticide: 1,2,3,4,5,6-$C_6H_6Cl_6$
Column: 2 x EnviroPrep, 300 x 25 mm (PL1210-6120EPA)
Eluent: Tetrahydrofuran
Flow Rate: 10 mL/min
Detector: RI

Ordering Information

Product	Part No.
EnviroPrep, 300 x 25 mm	PL1210-6120EPA
EnviroPrep, 150 x 25 mm	PL1210-3120EPA

For use with the EnviroPrep columns:

Product	Part No.
PLgel Prep Guard	PL1210-1120

Environmental and Food Clean-up Solutions from Varian

Varian offers a comprehensive product support solution for extraction, isolation, identification and quantitation in a wide range of environmental and food applications. Pesticide residues, PCBs, PAHs, toxins and antibiotic residues can be separated from foods (and packaging), plant or animal tissue, fish, soil and waste water.

Bond Elut™ SPE Cartridges

FactorFour™ GC Capillary Column

Pursuit™, Polaris™ and Pursuit XRs™ HPLC Columns

450-GC with 240-MS Ion Trap GC/MS

320-MS Quadrupole MS/MS with GC or LC Interface

Varian offers a complete support solution for the extraction, isolation, identification and quantitation of pesticide residues - request a copy of this brochure.

Complete Solutions for Pesticides Residue Analysis in Foods

GC or LC, coupled with mass spectrometry, are powerful tools for identifying and quantifying contaminants or adulterants in environmental samples.

VARIAN, INC.

EcoSpheres and EnviroPrep

SOLUTIONS FOR THE CLEAN-UP OF ENVIRONMENTAL & FOOD SAMPLES

Varian offers robust instrumentation, application-based consumables, and customer-focused services, backed by our global team of product and applications experts, ready to help you solve your analytical challenges. Whether you're monitoring impurities in drinking water, designing new therapeutic drugs, or developing cleaner fuels, our solutions deliver the sensitivity, flexibility and productivity your laboratory requires.

Varian Care Program

Our goal is to help you increase your productivity, maximize your uptime and achieve the highest return possible on your investment. Our experienced and highly-qualified support organization is strategically located throughout the world to ensure rapid response.

Be confident knowing that your instrument will deliver maximum performance, your users are fully trained and expert technicians will respond quickly to your support needs — because Varian cares.

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